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Effects of dispersed bonny light crude oil on two life stages of *Oreochromis niloticus*

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Abstract

The effects of Goldcrew, a chemical dispersant commonly used in crude oil spill incidents, to alter interactions between the components of the crude and the biota in the ecosystem, were studied. We used a static renewal bioassay to study its effects on Bonny Light crude oil using the tilapia, *O. niloticus* at various concentrations for 50 days. A control experiment was also set up. The exposure concentrations of the water accommodated fractions (WAF) of the mixture of the dispersant and Bonny Light crude, dispersed crude (DCOWAF-PAH) and the WAF of Bonny Light alone (COWAF-PAH) were 0.2, 0.4, 0.8 and 1.6 ml/L respectively. The mortalities of the fingerlings in the different concentrations of the DCOWAF-PAH were 66.7%, 23.3%, 86.75% and 100% in the 0.2, 0.4, 0.8 and 1.6 ml/L exposure concentrations respectively. At these concentrations, mortality was significantly different ($P < 0.05$) between the fingerlings and fry, with a total mortality of 69.2% among the fingerlings while the fry had 5.8% mortality in the 0.8 and 1.6 ml/L concentrations with 13.3% and 10.0% respectively but the differences were not statistically significant ($P < 0.05$). There was no mortality in the concentrations of the COWAF-PAH. It can be said that Bonny Light was made more toxic to *O. niloticus* by the presence of the dispersant; increased the susceptibility of the fingerlings to the mixture of crude and dispersant and influenced the behavior of the fish. There is therefore need to apply dispersants with caution especially around fish breeding and nursery grounds.

Keywords: Goldcrew, crude oil, *O. niloticus*.

Introduction

Dispersants are applied during oil-spill incidents to disperse the oil. They are specially designed for oil spill products that are composed of detergent-like surfactants in solvents that break the oil slick into small droplets. These droplets disperse into the water and are further broken down by natural processes. They also prevent the oil droplets from coming back together as another surface slick. Their effectiveness depends on the type of oil and the environmental conditions (National Oil and Hazardous Substances Response System, 2004). The environmental effects of the dispersants are influenced by physico-chemical and biological factors. The physico-chemical factors include: the quality of the dispersant, its concentration, temperature, salinity, oxygen, chemical stability and age of the test solution. The biological factors (exposed organisms) are: species, stage of development (eggs are most sensitive, while embryos are less sensitive than fry); seasonal variations, exposure history, acclimatization, health and nutritional status of fish.

The possible toxic effects of dispersed oil in the water column are of primary concern when assessing the use of dispersants to deal with an oil spill. The application of some dispersants on spilled oil has also been found to cause more damage to the living organisms inhabiting such ecosystem than the spilled oil acting singly (Otitoloju, 2005; Ogbonna et al., 2009). This study compared the sub-lethal effects of dispersed crude oil on two life stages (fry and fingerlings) of *O. niloticus*.

Materials and Methods

Three hundred and fifty fry and fingerlings of *O. niloticus* were procured from the African Regional Aquaculture Centre (ARAC) Aluu, Rivers State and kept in holding in plastic tanks (36×39×53cm of 25L) for fourteen days to acclimate in the laboratory of the Department of Animal and Environmental Biology, University of Port Harcourt using tap water. They were maintained with daily change of water to allow for stabilization before exposure and fed twice daily with Coppen feed of 0.8-1.2mm pellet size ad libitum at 5% body weight.

The Bonny Light crude oil and Goldcrew (SW) were procured from Nigerian Agip Oil Company in 1.5L air tight plastic bottles and stored at 28°C for preparation of the test solution. The water accommodated fractions (WAF) of the mixture of Bonny Light crude oil and Goldcrew (SW) dispersant (DCO_{WAF-PAH}) and that without Goldcrew (CO_{WAF-PAH}) were prepared under laboratory conditions (Reish and Oshida, 1986; Khan and Payne, 2005). The application ratio of crude oil to Goldcrew (SW) was determined to be 1:30, with a 1:30 dilution ratio of dispersant to distilled water. Range finding tests were done to determine the threshold concentrations (Lelei, 2007).

The static renewal bioassay of the two test exposures (DCO_{WAF-PAH} and CO_{WAF-PAH}) was conducted for fifty days with four concentrations each and three replicates with a factor of 0.2 that gave 0.2, 0.4, 0.8 and 1.6ml/L made up to 12L. Each tank had 10 test organisms for the tests. There was 12hr light and dark time respectively during exposures with a 48hr renewal of test media and mixtures to prevent sequestration (aging) by which chemicals tend to become less available with time for uptake by organisms for partitioning into the aqueous phase. The test organisms were observed for any abnormalities. One-factor ANOVA was used to test for differences among the concentrations, correlation analyses were done, statistical difference were accepted at $P < 0.05$. Mortality plot was extrapolated using probit values.

Results

The initial introduction of the toxicants into test media resulted in the restless of all fish with attempts to jump out of the tanks but they became calm within an hour. During exposure, the fry showed reduced startle response by the third day; and on the fourth day, reduced feeding was evident through feed waste until the eighth day. Startle response and feeding (reduced feed waste) were observed to improve by the tenth day, behaviour was normal by the twenty sixth day. Melanosis was observed by the fifth day in the fingerlings, narcosis with reduced startle response by the eighth day, reduced feeding and loss of scales were observed by the tenth day, mortality increased by the day in all concentrations till the sixteenth day but reduced by the nineteenth day when melanosis was no longer observed. In the 1.6ml/L concentration of DCO_{WAF-PAH}, all fingerlings died by the seventeenth day. Conversely, fish in CO_{WAF-PAH} showed no observable responses.

Table 1: Percentage mortalities of fingerlings exposed to sub-lethal concentrations of DCO_{WAF-PAH} for 50 days.

Concentrations (ml/L)	Number of test organisms	Mortalities with length of time of exposure in weeks							
		Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Total	% M
0.2	30	3	15	1	1	0	0	20	66.7
0.4	30	0	7	0	0	0	0	7	23.3
0.8	30	6	12	2	4	2	0	26	86.7
1.6	30	6	22	2	0	0	0	30	100
Total	120	15	56	5	5	2	0	83	69.2

Table 2: Mortalities and percentages of fry of *O. niloticus* exposed to sub-lethal concentrations of DCO_{WAF-PAH} for 50 days.

Concentrations (ml/L)	Number of test organisms	Mortalities with length of Time of Exposure in weeks								
		Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Total	%M
0	10	—	—	—	—	—	—	—	—	—
0.1	30	—	—	—	—	—	—	—	—	—
0.4	30	—	—	—	—	—	—	—	—	—
0.8	30	2	—	2	—	—	—	—	4	13.3
1.6	30	—	—	—	—	—	—	3	3	10.0
Total	120	2	—	2	—	—	—	3	7	5.8

The mortality profiles among fingerlings exposed to DCO_{WAF-PAH} were 12.55%, 59.2, 63.3%, 67.5%, 69.2% and 69.2% in the 1st to 6th weeks respectively (Table 1). The percentage mortality in the different exposure concentrations were 66.7% (20), 23.3% (7), 86.75% (26) and 100% (30) for 0.2, 0.4, 0.8 and 1.6ml/L respectively. Mortality increased with increase in concentration. Among the fry, (Table 2), the total mortality under DCO_{WAF-PAH} was 5.8% (7) at the end of the exposure period (week7) with 1.7% (3), and 3.4% (4) by the end of week 1 and week 2 respectively. Mortality levels under 0.8 and 1.6ml/L exposure concentrations were 13.3% (4) and 10.0% (3) respectively. Conversely, there was no mortality in the CO_{WAF-PAH} (Table 3) irrespective of concentration.

Table 3: Mortalities and percentages of post-hatchlings and fry of *O. niloticus* exposed to sub-lethal concentrations of $CO_{WAF-PAH}$ for 50 days.

Concentrations (ml/L)	Number of test organisms	Mortalities with length of time of exposure in weeks								
		Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Total	%M
0	10	—	—	—	—	—	—	—	—	—
0.1	30	—	—	—	—	—	—	—	—	—
0.4	30	—	—	—	—	—	—	—	—	—
0.8	30	—	—	—	—	—	—	—	—	—
1.6	30	—	—	—	—	—	—	—	—	—
Total	120	0	0	0	0	0	0	0	0	0

ANOVA results for the fingerlings, showed that mortalities were significantly different ($P < 0.05$) between (time) and within (concentrations) with significant differences in week 1, 3, 4, 5 and 6 but not significant ($P < 0.05$) for week 2. There was positive correlation for week 1, 2, 4 and 6 but negative correlation for weeks 3 and 5 of time against concentration. For the fry, the ANOVA showed that the mortalities were not significant ($P < 0.05$) between (time) and within (concentration). Weeks 1 and 3 had positive correlation for time against concentration. The $CO_{WAF-PAH}$ at different concentrations recorded no mortality.

Probit plot was used to determine the time taken for fifty percent mortality to occur in 50-day for the fingerlings in the concentrations of $DCO_{WAF-PAH}$. This was determined to be 1 week, 3 days, 15 hours which implied that it took about 11 for 50% (69.2) mortality to occur. This was not the case with the fry as only 5.8% mortality was recorded during the fifty day exposure period.

Discussion

Behavioural responses observed in the exposed fish were influenced by the exposure regime. The reduced startle responses, narcosis, and poor feeding observed during the early exposure period were in line with other findings (Ramachandran et al., 2004; Khan and Payne, 2005). The fry showed better responses than the fingerlings indicating that fingerlings had lower resistance than the fry to the exposure of the $DCO_{WAF-PAH}$. This is attributable to increased susceptibility of fish to toxicants with stage of development the chorion conferred protection on the early stages of fish in line with previous findings (Pollino and Holdaway, 2002).

Mortalities in the different exposure regimes showed that mortality increased with increase in concentration as observed in the percentage mortalities. The strength of this finding was shown by the positive correlation between the concentrations of the exposure chemicals with time as against the control which recorded no mortality. This affirmed the fact that the chemical mixtures were the cause of the observed mortality.

The $DCO_{WAF-PAH}$ was implicated for being more toxic on fingerlings as against the $CO_{WAF-PAH}$ that had no mortality among the fry. This implied complicity of the dispersant in the increased mortality of the fingerlings confirmed the age-susceptibility relationship in organisms exposed to toxicants. This study has confirmed that the effects of the use of dispersants can have serious effects even at very low concentrations especially on fingerlings of this tilapia.

Table 3: Mortalities and percentages of post hatchlings and fry of *O. niloticus* exposed to sub-lethal concentrations of $CO_{WAF-PAH}$ for fifty days.

Concentrations (ml/L)	Number of test organisms	Mortalities with length of time of exposure in weeks								
		Week 1	Week 2	Week 3	Week 4	Week 5	Week 6	Week 7	Total	%M
0	10	—	—	—	—	—	—	—	—	—
0.1	30	—	—	—	—	—	—	—	—	—
0.4	30	—	—	—	—	—	—	—	—	—
0.8	30	—	—	—	—	—	—	—	—	—
1.6	30	—	—	—	—	—	—	—	—	—
TOTAL	120	0	0	0	0	0	0	0	0	0

Conclusion

The impacts of toxicants on the biological components of ecosystems are the results of a cascade of events that begin with their introduction into the environment, their assimilation or uptake by impacted organisms, and with their interactions with the biological components. The physiological responses to these interactions are made manifest in the behavioural, metabolic, developmental and reproductive aspects of the affected organisms. The climax of these interactions may ultimately lead to death. These would eventually affect population/population dynamics especially of year classes since susceptibility increased with age.

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